

CGCCTGTGCCCTCTGCCTGGGAGCCTGGGGCCGCCTGTCTGCGCGGTCCGGATGCGCTCAGGTCAAGGTTCCCTTTTCG
CGGCTGTCTCCCAAGCCCCTAACTAGTGACTTCCACTGTGGCGGGCAGGGAAGCCATTGGCAGAACCTAGCCAGTCA
GGAATCTGCATCTCTTCCCTCATTATCCTCTCCCTGGCATTGCTTTGCTCGGGTCCAGCTCAGTTGGTGACGCGGCC
CCTTCTCCCCAGGTTGGGATCCACGGAAGCAGGGGTGCAGGCCGGCCAGGCACTGTGCCATGAGCCAGAGCCCCGAGG
TTCGTGACCCGGAGGGGCGGCTCTCTAAAGGCTGCCCCTGGAGCCGGCACC CGCGCAACGAGAGCCAGGACTATTT
GCTGATGGACGAGCTGGGAGACGACGGCTACCCGCAGCTCCCGCTGCCACCGTATGGCTACTACCCAGCTTCCGGG
GTAATGAAAACAGACTGACTCACCGGCGGCAGACGATTCTTCGTGAGAAGGGAAGAAGGTTAGCTAATCGAGGACCA
GCATACATGTTTAAATGATCATTCAACAAGCCTGTCTATTGAGGAAGAACGCTTTCTAGATGCGAGTTGAATATGGCAA
CATCCCAGTGGTCTGGAAGATGCTAGAAGAGTGTCAATCCCTCAATGTTAACTGTGTGGATTACATGGGCCAGAATG
CCCTACAGCTGGCTGTGGCCAATGAGCACTTGGAAATCACAGAGCTGCTACTCAAGAAGGAAAACTTGTCTCGAGTT
GGGGATGCTTTACTTTTAGCCATTAGTAAAGGTTATGTACGGATTGTGGAGGCAATCCTCAACCATCCATCTTTTGC
TGAAGGCAAAAGGTTAGCGACAAGCCCCAGCCAGTCTGAACCTCAGCAAGATGACTTTTATGCCTATGATGAAGATG
GGACGCGGTTCTCCCATGATGTGACTCCAATCATTCTCGCTGCACATTGCCAGGAATATGAAATTGTGCATACCCCTC
CTGAGAAAAGGGTGCCCGATTGAGCGGCCTCATGATTACTTCTGCAAGTGTACAGAAATGCAGCCAGAAGCAGAAGCA
TGATTCCCTTCAGCCACTCTAGATCCAGGATCAATGCATACAAAGGTCTGGCAAGTCCAGCATACTGTCAATTGTCCA
GTGAAGATCCAGTCATGACTGCTTTAGAACTTAGCAATGAGCTGGCAGTGCTTGCCAACATTGAGAAAAGAGTTCAAG
AATGACTACAGGAAGCTGTCTATGCAGTGCAAGGATTTCTGTTGTTGGTCTCTTGGACCTCTGCAGAAAACACAGAGGA
AGTGAGAGGCCATCCTGAATGGGGATGCAGAGACTCGCCAGCCCGGGGACTTCGGCCGTCCAAATCTCAGCCGTTTAA
AACTTGCTATTAAGGATGAAGTAAAAAAATTTGTGGCTCATCCAACTGTCAGCAACAGCTCCTGTCCATATGGTAT
GAGAACCTCTCTGGTTTACGGCAGCAGACCATGGCAGTGAAGTTCCTCGTGGTCTTGTGCTGTTGCCATTGGATTGCC
CTTCTCGGCTCTCATATACTGGTGTGCTCCTTGCAAGATGGGGAAGATATTGCCGAGACCGTTTATGAAGTTTG
TAGCACACGCAGCCTCCTTACCATTTCCTGGGGCTGCTCGTCATGAATGCAGCTGACAGATTTGAAGGCACCAAG
CTCCTCCCTAATGAAACCAGCACAGATAATGCAAGGCAGCTGTTTCAGGATGAAAACATCCTGTTTCTCATGGATGGA
GATGCTCATTATATCCTGGGTAATAGGCATGATATGGGCTGAATGTAAAGAAATCTGGACTCAAGGCCCCAAAGAAT
ACTTATTTGAGTTGTGGAATATGCTTGACTTTGGAATGCTGGCAATCTTTGCAGCATCATTCAATTGCAAGATTTATG
CGGTTCTGGCATGCATCCAAAGCTCAGAGCATCATTGATGCAATGATACTTTAAAGGATTTGACAAAAGTCACACT
GGGGGACAACGTTAAATACTACAATCTGGCCAGGATAAAGTGGGACCCTACTGATCCTCAGATCATCTCTGAAGGTC
TTTATGCAATCGCTGTGGTTTTTAAGTTTTCTCCAGAATAGCTTACATTTTACCAGCAAATGAAAGCTTTGGACCTCTG
CAGATTTCACTTGGAAGAACAGTGAAAGATATCTTCAAATTCATGGTCATATTTCATCATGGTGTTTGTAGCCTTTAT
GATTGGAATGTTCAACCTTTACTCCTACTACATTGGCGCAAAACAGAATGAAGCATTACACAACAGTTGAGGAAAAGTT
TTAAGACACTGTTCTGGGCTATCTTTGGTCTTTCTGAAGTGAAGTCAGTGGTCATTAACTACAATCACAAGTTCATT
GAAAACATCGGCTACGTTCTGTATGGTGTCTATAATGTCACAATGGTCATTGTTTTGCTAAATATGTTAATTGCGAT
GATCAATAGTTTCAATCCAGGAAATTGAGGATGATGCGGACGTGGAGTGGAAAGTTTGCAAGGGCCAAATGTGGTTTT
CCTACTTTGAGGAGGGGAGAACACTTCCTGTCCCCTTCAATCTTGTACCAAGTCCAAAATCCTTGCTTTATCTCCTA
TTGAAATTTAAGAAATGGATGTGTGAGCTCATCCAGGGTCAAAGCAAGGCTTCCAAGAAGATGCAGAGATGAACAA
GAGAAATGAAGAAAAGAAATTTGGAATTTAGGAAGTCACGAAGACCTTTCAAAATTTTCACTTGACAAAAATCAGT
TGGCACACAACAAACAATCAAGTACAAGGAGCTCAGAAGATTATCATTTAAATAGTTTCAGTAACCTTCCAAGACAA
TATCAGAAAATCATGAAGAGACTCATTAAAAGATATGTATTGCAGGCCAGATTGATAAGGAGAGCGATGAGGTGAA
TGAAGGGGAATTGAAGGAAATTAAGCAAGACATCTCAAGTCTCCGTTATGAACTCCTTGAAGAGAAAATCACAGAACT
CAGAAGACCTAGCAGAGCTCATTAGAAAACCGGGGAGAGACTGTGTTAGAGCCAAAGCTGGAGGAAAGCCGCAGA
TAGAGCAGAGCCCCCTCAGAAGTGCATATTTATTTCTCCACTTGAAGCCATATTATTTTCTGACTTATTTTAAAGT
GTCAATGATAAAAAGTATGTTAACTGATAACTTGGATCATTAGAGTCCTAATATCAAGCTTTTGGGAGATTAAAT
TGCATTGCTGAGGGCTAACAATTGCTG (SEQ ID NO:1)

FIGURE 1

MSQSPRFVTRRGGSLKAAPGAGTRRNESQDYLLMDELGDDGYPQLPLPPYGYYPSPFRGNENRLTHRRQTI
LREKGRRLANRGPAYMFNDHSTSLSEERFLDAVEYGNIPVWKMLEECHSLNVNCVDYMGQNALQLAV
ANEHLEITELLKKENLSRVGDALLLAISKGYVRIVEAILNHPSFAEGKRLATSPSQSELQQDDFYAYDE
DGTRFSDVTPILAAHCQEYEIVHTLLRKGARIERPHDYFCKCTECSQKQKHDSFSHSRSRINAYKGLA
SPAYLSLSSDPVMTALELSNELAVLANIEKEFKNDYRKLSMQCKDFVVGLLDLCRNTEEEVEAILNGDAE
TRQPGDFGRPNLSRLKLAIKDEVKKFVAHPNCQQQLLSIWYENLSGLRQQTMVAVKFLVVLAVAIGLPFLA
LIYWCAPCSKMGKILPRPFMKFVAHAASFTIFLGLLVMNAADRFEGTKLLPNETSTDNARQLFRMKTSCF
SWMEMLIISWVIGMIWAECKEIIWTQGPKEYLFELWNMLDFGMLAIFAASFIARFMAFWHASKAQSIIDAN
DTLKDLTKVTLGDNVKYYNLARIKWDPTDPQIISEGLYAIIVVLSFSRIAYILPANESFGPLQISLGRTV
KDIFKFMVIFIMVFVAFMIGMFNLYSYYIGAKQNEAFTTVEESFKTLFWAIFGLSEVKSVVINYNHKFIE
NIGYVLYGVYNVTMIVVLLNMLIAMINSSFQEI EDDADVEWKFARAKLWFSYFEEGRTPVPFNLVPSPK
SLLYLLLKFKKWMCELIQGQKQGFQEDAEMNKRNEEKKFGISGSHEDLSKFSLDKNQLAHNKQSSTRSSE
DYHLNSFSNPPRQYQKIMKRLIKRYVLQAQIDKESDEVNEGELKEIKQDISSRLRYELLEKSQNSD LAE
LIRKLGERLSLEPKLEESRR (SEQ ID NO:2)

FIGURE 2

underlined = deleted in targeting construct

BOLD = sequence flanking Neo insert in targeting construct

CGCCTGTGCCCTCTGCCTGGGAGCCTGGGGCCGCTGTCTGCGCGGTCCGGATGCGCTCAGGTCAAGGTTCCCT
 TTCGCGGCTGTCTCCCAAGCCCCTAACTAGTGACTTCCACTGTGGCGGGCAGGGAAGCCATTGGCAGAACCTA
 GCCAGTCAGGAATCTGCATCTCTTCCCTCATTATCCTCTCCCTGGCATTGCTTTGCTCGGGTCCAGCTCAGTT
 GGTGACGCGGCCCCCTTCTCCCCAGGTTGGGATCCACGGAAGCAGGGGTGCAGGCCGGCCAGGCACTGTGCCAT
 GAGCCAGAGCCCCGAGGTTCTGTGACCCGGAGGGGCGGCTCTCTAAAGGCTGCCCTGGAGCCGGCACCCGGCGC
 AACGAGAGCCAGGACTATTTGCTGATGGACGAGCTGGGAGACGACGGCTACCCGAGCTCCCGTGGCCACCGT
 ATGGCTACTACCCAGCTTCCGGGGTAATGAAAACAGACTGACTCACCGGCGGCAGACGATTCTTCGTGAGAA
 GGAAGAAGGTTAGCTAATCGAGGACCAGCATACATGTTTAAATGATCATTCAACAAGCCTGTCTATTGAGGAA
 GAACGCTTTCTAGATGCAGTTGAATATGGCAACATCCAGTGGTCTGGAAGATGCTAGAAGAGTGTCTATTCCC
 TCAATGTTAACTGTGTGGATTACATGGGCCAGAATGCCCTACAGCTGGCTGTGGCCAATGAGCACTTGGAAAT
 CACAGAGCTGCTACTCAAGAAGGAAAACTTGTCTCGAGTTGGGGATGCTTTACTTTTAGCCATTAGTAAAGGT
 TATGTACGGATTGTGGAGCAATCCTCAACCATCCATCTTTTGTGTAAGGCAAAAGGTTAGCGACAAGCCCCA
 GCCAGTCTGAACCTCAGCAAGATGACTTTTATGCCTATGATGAAGATGGGACGCGGTTCTCCCATGATGTGAC
 TCCAATCATTCTCGTGCACATTGCCAGGAATATGAAATTGTGCATACCCTCCTGAGAAAGGGTGCCCGGATT
 GAGCGGCTCATGATTACTTCTGCAAGTGACAGAATGCAGCCAGAAGCAGAAGCATGATTCCCTTCAGCCACT
 CTAGATCCAGGATCAATGCATACAAAGGTCTGGCAAGTCCAGCATACCTGTCTATTGTCCAGTGAAGATCCAGT
 CATGACTGCTTTAGAAGTTAGCAATGAGCTGGCAGTGCTTGCCAACATTGAGAAAGAGTTCAAGAATGACTAC
 AGGAAGCTGTCTATGCAGTGCAAGGATTTCTGTTGTTGCTCTTGGACCTCTGCAGAAACACAGAGGAGTGG
 AGGCCATCCTGAATGGGGATGCAGAGACTCGCCAGCCCGGGGACTTCGGCCGTCCAAATCTCAGCCGTTTAA
 ACTTGCTATTAAGGATGAAGTAAAAAATTTGTGGCTCATCCAACTGT**CAGCAACAGCTCCTGTCCATATGG**
TATGAGAACCTCTCTGGTTTACGGCAGCAGACCATGGCAGTGAAGTTCTCGTGGTCTCTGCTGTTGCCATTG
GATTGCCCTTCTCGGCTCTCATATACTGGTGTGCTCCTTGAGCAAGATGGGGAAGATATTGCCGAGACCGTT
 CATGAAGTTTGTAGCACACGCAGCCTCCTTACCATTCTTCTGGGGCTGCTCGTCATGAATGCAGCTGACAGA
 TTTGAAGGCACCAAGCTCCTCCCTAATGAAACCAGCACAGATAATGCAAGCAGCTGTTCCAGGATGAAAACAT
 CCTGTTTCTCATGGATGGAGATGCTCATTATATCCTGGGTAATAGGCATGATATGGGCTGAATGTAAAGAAAT
 CTGGACTCAAGGCCCCAAAGAATACTTATTTGAGTTGTGGAATATGCTTGACTTTGGAATGCTGGCAATCTTT
 GCAGCATCATTATTGCAAGATTTATGGCGTTCTGGCATGCATCCAAAGCTCAGAGCATCATTGATGCAATG
 ATACTTTAAAGGATTTGACAAAAGTCACACTGGGGGACAACGTTAAATACTACAATCTGGCCAGGATAAAGTG
 GGACCCTACTGATCCTCAGATCATCTCTGAAGGTCTTTATGCAATCGCTGTGGTTTTAAGTTTTCTCCAGAATA
 GCTTACATTTTACCAGCAAAAGAAAGCTTTGGACCTCTGCAGATTTCACTTGGAAGAAGTGAAGATATCT
 TCAAATTCATGGTCATATTCATCATGGTGTGTTGTAGCCTTTATGATTGGAATGTTCAACCTTTACTCCTACTA
 CATTGGCGCAAAACAGAAATGAAGCATTACACAACAGTTGAGGAAAGTTTTAAGACACTGTTCTGGGCTATCTTT
 GGTCTTTCTGAAGTGAAGTCAGTGGTCATTAATACTACAATCACAAGTTCATTGAAAACATCGGCTACGTTCTGT
 ATGGTGTCTATAATGTCACAATGGTCATTGTTTTGCTAAATATGTTAATTGCGATGATCAATAGTTCATTCCA
 GGAAATTGAGGATGATGCGGACGTGGAGTGAAGTTTGCAAGGGCCAAATTTGGTTTTCTACTTTGAGGAG
 GGGAGAACAATCTCTGCCCCCTTCAATCTTGTAACCAAGTCCAAATCCTTGCTTTATCTCCTATTGAAATTTA
 AGAAATGGATGTGTGAGCTCATCCAGGCTCAAAAGCAAGGCTTCCAAGAAGATGCAGAGATGAACAAGAGAAA
 TGAAGAAAAGAAATTTGGAATTTTCAAGGAGTCAAGAGACCTTTCAAATTTTCACTTGACAAAAATCAGTTG
 GCACACAACAAACAAATCAAGTACAAGGAGCTCAGAAGATATCATTAAATAGTTTCAGTAACCCCTCCAAGAC
 AATATCAGAAAATCATGAAGAGACTCATTAAAAAGATATGATTGCAGGCCAGATTGATAAGGAGAGCGATGA
 GGTGAATGAAGGGGAATTGAAGGAAATTAAGCAAGACATCTCAAGTCTCCGTTATGAACTCCTTGAAGAGAAA
 TCACAGAACTCAGAAGACCTAGCAGAGCTCATTAGAAAACCGGGGAGAGACTGTGCTTAGAGCCAAAGCTGG
 AGGAAAGCCGCAGATAGAGCAGAGCCCTCAGAAGTGCATATTTATTTCTCCACTTGAAGCCATATTATTTTC
 TGAATTTTAAAGTGTCAATGATAAAAAGTATGTTAACTGATAAAGTGGATCATTAGAGTCCATAATAT
 CAAGCTTTTGGGAGATTAAATTGCATTGCTGAGGGCTAACAATTGCTG

FIGURE 3

Gene Sequence Structure *

1497 bp

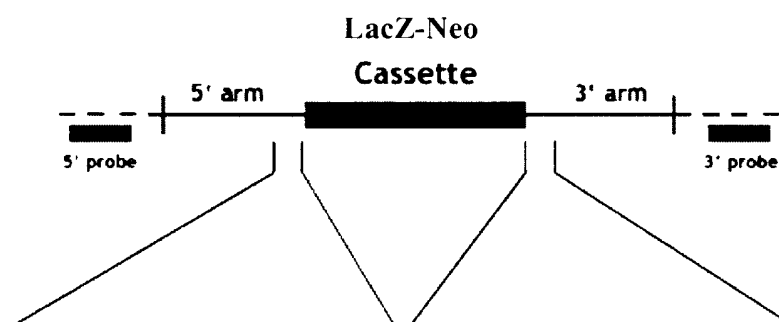
Sequence Deleted

1509 bp

Size of full-length
cDNA: 3261 bp

Targeting Vector*
(genomic sequence)

Arm Length:
5': 3.5 kb
3': 1.5 kb



————— Targeting Vector
- - - - - Endogenous Locus

* Not drawn to scale

5' > TCCTCAATTCTAACTGCATTT
CTTCTGGAAAAGAATAAAACGATT
CACCAGAGCTCCAGAGGATAGCCT
AAGCTGAGTTGTTTTAATCAAAT
CATTCTGTGTGCTGTCTCACCCCT
AGTTTGTGGCTCATCCAAGCTGTC
AGCAACAGCTCCTGTCCATATGGT
ATGAGAACCTCTCTGGTTTACGGC
AGCAGACCATG<3'
(SEQ ID NO:3)

5' > TCGTGGTCCTTGCTGTTGCCA
TTGGATTGCCCTTCCTGGCTCTCA
TATACTGGTGTGCTCCTTGCAGCA
AGGTATGTCTGTGAGTCCTGCAGT
CCATCTGTAGTTGAATTCTGTCCA
GCAGGCAAAGATCTAGCTCCAAAA
TGAAAAATATGATTTGAAGTACACA
GGTTCACATAATCTTTCTATTGT
TTGAGAATTTTC<3'
(SEQ ID NO:4)

FIGURE 4

5/5

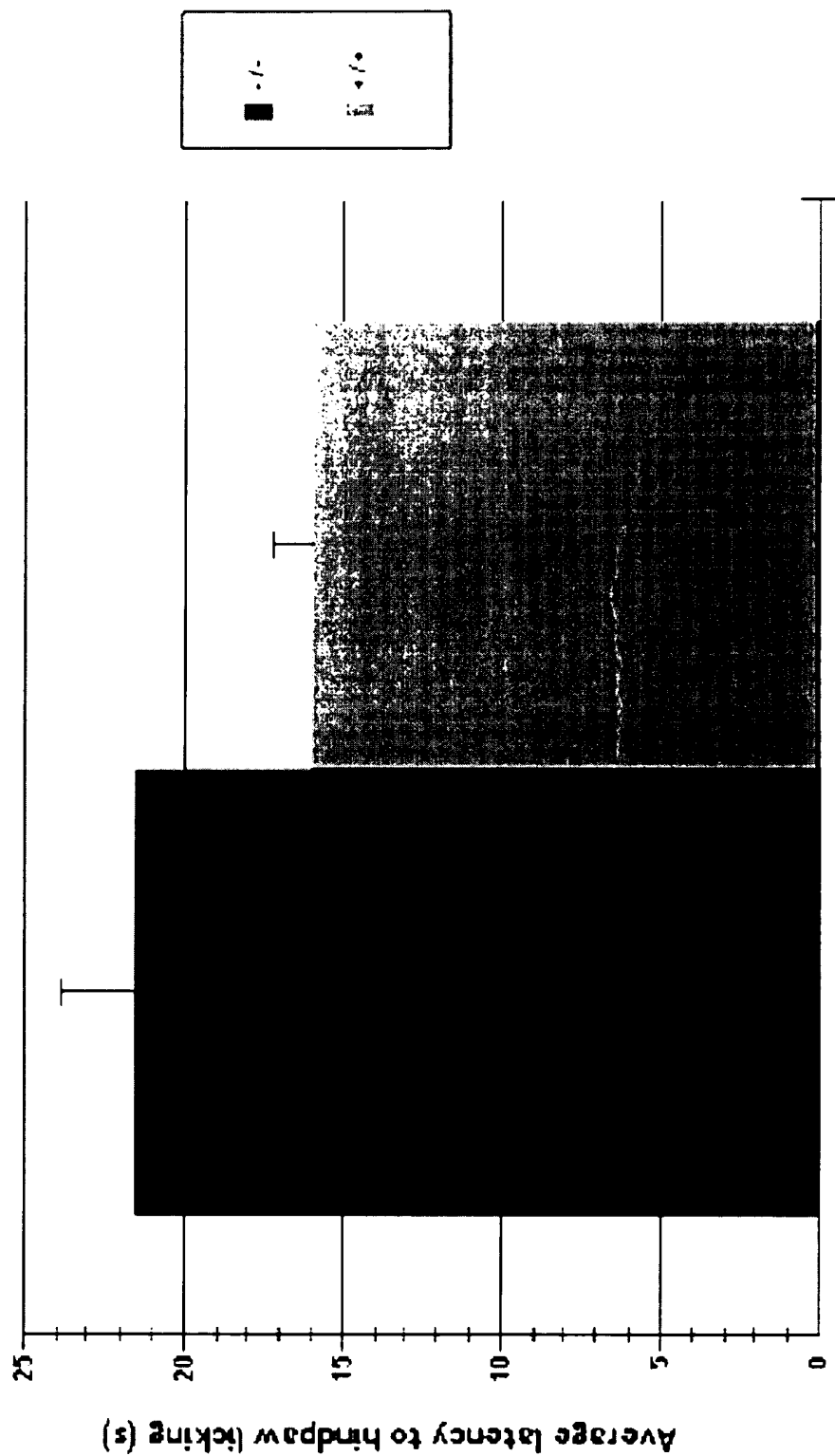


FIGURE 5